Host-Foraging Success of Three Species of *Trichogramma* (Hymenoptera: Trichogrammatidae) in a Simulated Retail Environment

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ABSTRACT   Three species of trichogrammatid egg parasitoids (*Trichogramma deion* Pinto & Oatman, *Trichogramma ostriniae* Pang & Chen, and *Trichogramma pretiosum* Riley) (Hymenoptera: Trichogrammatidae) were evaluated under laboratory conditions as potential biological control agents for the Indianmeal moth, *Plodia interpunctella* (Hübner) (Lepidoptera: Pyralidae), on retail shelves. A single shelving unit was used in each trial and a grid of sentinel egg disks was used to evaluate foraging success. The shelving consisted of pallet units with five shelves that were either bare or stocked with empty cereal boxes. In each replicate, ≈500 female *Trichogramma* were released at the center of the shelving unit and allowed to forage for 48 h. Percentage of egg parasitism and percentage of host egg mortality were recorded after 7 d. Foraging success as well as the spatial pattern of parasitism differed significantly among the three *Trichogramma* species. Percentage of egg parasitism was ≈4 times greater for *T. deion* than for *T. ostriniae* or *T. pretiosum*. The vertical distribution of parasitism by *T. deion* was also more uniform than for the other two species. In addition, the presence of packaging affected the foraging efficiency of *T. ostriniae* and *T. pretiosum* but not *T. deion*. Based on these findings, *Trichogramma deion* may be the best-suited candidate for augmentative biological control of *P. interpunctella* in retail stores, and a central release point of *T. deion* will likely provide adequate coverage of products on pallet-type shelving.

KEY WORDS   habitat complexity, stored-products, augmentative biological control, retail stores

The Indianmeal moth, *Plodia interpunctella* (Hübner) (Lepidoptera: Pyralidae), is a ubiquitous pest of finished stored-products in North America. It is capable of infesting a broad range of products, including raw and processed cereal products, animal feed, dried fruit, nuts, and garlic (Cox and Bell 1991, Perez-Mendoza and Aguilera-Peña 2004). In retail stores, damage can result in customer complaints and a subsequent loss of business due to infested packaged products. Traditionally, management of the Indianmeal moth in retail stores has consisted of either sanitation or application of insecticidal fogs, fumigants, and sprays to infested areas (Cox and Bell 1991). In recent years, regulatory constraints, such as the Food Quality and Protection Act, and consumer concern over pesticide

residues have limited the availability of insecticides for use in stored products and complicated their use (Arthur and Rogers 2003). Biological control of *P. interpunctella* by using natural enemies such as egg parasitoids in the genus *Trichogramma* (Hymenoptera: Trichogrammatidae) is one possible pest management alternative.

Species of *Trichogramma* have had a long history of use as biological control agents of lepidopterous pests on field crops and in greenhouses (Keller et al. 1985, Li 1994). *Trichogramma* spp. also have been evaluated against a variety of stored product moths in bulk peanut storage (Brower 1988), bulk wheat storage (Schöller et al. 1994, 1996), and bakeries (Prozell and Schöller 1994, Steidle et al. 2001) as well as in warehouses and retail stores in Europe (Prozell et al. 1996). Stored-product moths commonly oviposit on packages, and on shelves holding stored-product packages. *Trichogramma* spp. are especially promising as biological control agents on finished products because they attack the egg stage of the pests, thereby preventing the invasion of products by first instars.

The selection of *Trichogramma* species and strains that are the most suitable for a particular habitat has been identified as a priority for successful biological control (Schöller et al. 1996). Evaluating host-foraging efficiency is one means of assessing suitability of dif-
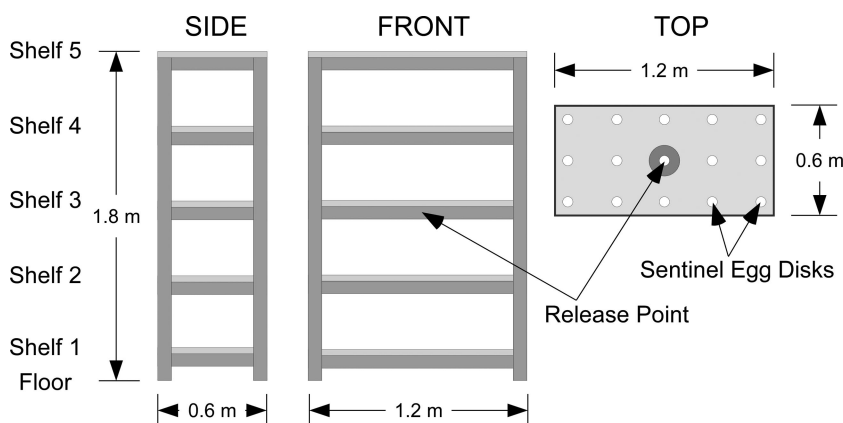
Fig. 1. Diagram of pallet shelving unit showing 3 by 5 sentinel egg disk layout and the central release point for *Trichogramma* species. Sentinel egg disks were spaced in a 33- by 28.6-cm grid.

ferent species or strains. Because *Trichogramma* are most commonly released in large numbers from one or more release points, rather than distributed evenly across the targeted area, the impact of habitat complexity on *Trichogramma* dispersal and host finding is an especially important consideration. For example, a number of recent studies have demonstrated the potential for habitat complexity to mitigate host-foraging by *Trichogramma* in the field (McCravy and Berisford 1998), under greenhouse conditions (Gingras et al. 2003), and in the laboratory (Andow and Prokrym 1990, Lukianchuk and Smith 1997, Gingras et al. 2002, Gingras and Boivin 2002, Andow and Olson 2003). However, none of these studies were conducted in retail stores or warehouses, where habitat complexity is a function of shelving type and the amount of packaged goods. Therefore, the objective of this study was to compare the abilities of three species of *Trichogramma* to disperse among shelves and parasitize Indianmeal moth eggs in a simulated retail store environment in the presence and absence of packaging. Based on results from a previous study (Schöller and Fields 2003), the three species selected for comparison were *Trichogramma deion* Pinto & Oatman, *Trichogramma ostriniae* Pang & Chen, and *Trichogramma pretiosum* Riley.

Materials and Methods

Insects. All host and parasitoid species were reared at the USDA–ARS Grain Marketing and Production Research Center (GMPRC) in Manhattan, KS. Colonies of *T. deion*, *T. pretiosum*, and Mediterranean flour moth, *Ephestia kuehniella* Zeller (Lepidoptera: Pyralidae), were originally obtained from Beneficial Insectaries (Redding, CA) in March, 2004, April 2003, and February, 2002, respectively (Kansas State University Entomology Voucher 171). *T. ostrinae* were obtained from a colony maintained by Dr. Michael Hoffman, Cornell University (Ithaca, NY) in November 2003 (Kansas State University Entomology Voucher 171). *P. interpunctella* served as the experimental host for *Tri-*

chogramma and was obtained from a laboratory colony initiated at the GMPRC in December 2001 and maintained continuously on a standard diet of cracked wheat, wheat shorts, honey, water, and glycerin (McGaughey and Beeman 1988) and under the environmental conditions described below for *Trichogramma*. All three species of *Trichogramma* were reared in growth chambers set at $26 \pm 1°C$, $60 \pm 5\%$ RH, and a photoperiod of 16:8 (L:D) h. Hosts used for the *Trichogramma* colonies were eggs of *E. kuehniella* that had been sterilized with UV light (6-W Spectronics BLE 6254s) for 1 min (eggs were placed ≈2.5 cm below the bulb).

Experimental Design. Experiments were conducted between 2 July 2003 and 27 July 2004 in walk-in growth chambers ($23 \pm 1°C$ and $45 \pm 5\%$ RH with a photoperiod of 16:8 [L:D] h) at the GMPRC. Environmental conditions were selected based on mean values of data collected over a 6-mo period from April to November 2002 in four retail stores in Manhattan, KS. Sentinel egg disks were used to map parasitism. These consisted of 1-cm-diameter disks punched from cardstock to which four *P. interpunctella* eggs (<18 h old) were attached using an inert, plant-based glue, Traganth (Merck, Whitehouse Station, NJ).

Trials were conducted for 48 h on five-tiered open shelving units measuring 1.2 by 0.6 by 1.8 m (length by width by height), either with or without packaging. Shelving unit risers were constructed of painted sheet metal with shelves made of particleboard. Total surface area of one shelf was ≈7.92 m². Sentinel egg disks were evenly distributed in a 3 by 5 configuration over each shelf and on the floor directly beneath the shelving unit, with the exception of the third shelf from the bottom, which had the center egg disk replaced with the *Trichogramma* release point (Fig. 1). The resulting three-dimensional grid consisted of six tiers of 15 disks per shelf with the third (central) shelf having 14 disks. Sentinel eggs were spaced in a grid measuring 33 by 28.6 cm and extending across the entire shelf. For each replicate, an additional 30–40 *P. interpunctella* egg disks were kept in sealed petri dishes, on an adjacent

shelving unit, within the experimental growth chamber. These eggs served as a control for mortality from sources other than *Trichogramma.* Replicates in which >20% control egg mortality was observed were omitted from the experiment. Replicates were run at least 5 d apart to ensure that any *Trichogramma* surviving from preceding replicates had died.

We used a 2 by 3 factorial design, with two levels of shelving (either empty or with packaging) and three levels for species (three different *Trichogramma* species tested). Each of the six species × shelving treatment combinations was replicated five times for a total of 30 experimental units. *T. deion* trials were run between 2 July 2003 and 20 October 2003, trials for *T. ostriniae* were run between 22 December 2003 and 21 July 2004, and trials for *T. pretiosum* were run between 6 June 2004 and 27 July 2004. In treatments that included packaging, an assortment of empty cereal boxes was used. Boxes were placed on the shelves in between sentinel egg disks in four columns, with six boxes per column and boxes spaced approximately equidistant to fill the width of the shelf. Total surface area of packages per shelf was 3.97 m^2.

For each replication of the experiment, *Trichogramma* spp. were released by placing parasitized host eggs containing late pupal stages. Releases were scheduled so that ≈500 adult female parasitoids would emerge within 1 h after placement. The number of parasitized eggs per release was varied among species to compensate for sex ratio differences determined in previous generations. *T. deion*, *T. ostriniae*, and *T. pretiosum* had female:male sex ratios of 2:1, 3:1, and 49:1, respectively, resulting in releases of 750, 667, and 510 parasitized eggs, respectively.

Data Collection. Sentinel egg disks were labeled and collected after 48 h and held for 7 d in a growth chamber set for 26 ± 1°C and 60 ± 5% RH, at which point the eggs on individual cards were observed at 160–400× magnification by using a stereomicroscope. Data collected included numbers of eggs hatched, parasitized, or dead. Egg state was visually determined with eggs that were empty graded as "hatched," eggs containing pupal *Trichogramma* graded as "parasitized," and eggs containing unidentifiable substances as "dead." Parasitism was easily distinguished by a darkened appearance of the egg and/or the presence of reddish eyespots. Data were used to calculate the percentage of parasitized eggs per shelf (averaged over all sentinel eggs on each shelf), the percentage of egg cards per shelf that had any parasitism, and total mortality related to parasitism (parasitized eggs + eggs graded as dead). The latter value was corrected for mortality unrelated to parasitism by using Abbott's formula (Abbott 1925): corrected mortality = (observed total mortality − control mortality)/(1 − control mortality).

Data Analysis. Data were analyzed using the MIXED, GLM, and FREQ procedures (SAS Institute 2000). A complete factorial mixed model analysis of variance (ANOVA) (MIXED procedure) was run using species and packaging nested within experimental run as the random factor and species, packaging, and shelf level as fixed factors. Degrees of freedom were

corrected using the Kenward–Rodgers correction. An LSMEANS statement was used to calculate individual level comparisons by *t*-tests with *P* values corrected using the TUKEY option. Six additional one-way ANOVA models (GLM procedure) were run to compare differences among shelving levels by packaging treatment and species, with multiple comparisons computed using Tukey's honestly significant difference (HSD) test. Finally, a mixed model *t*-test was run for each species, comparing percentage of parasitism, percentage of patches attacked, and percentage of corrected total mortality between packaging treatments for each shelf level. Packaging nested within experimental run was used as the random factor to account for potential time effects. All percentages were adjusted using the arcsine square-root transformation [arcsine(x$^{0.5}$)]. The dispersion of parasitism across the six levels (five shelves and the floor) of sentinel egg disk grids was tested to see whether it fit a uniform distribution by using a chi-square goodness-of-fit-test, Proc FREQ, (SAS Institute 2000).

Results

Percentage of Parasitism. Significant treatment effects and interactions were detected for percentage of parasitism, including species ($F = 78.54$; df = 2, 24; $P < 0.01$), shelf level ($F = 25.92$; df = 5, 120; $P < 0.01$), shelf level and packaging ($F = 2.46$; df = 5, 120; $P = 0.04$), and species and shelf level ($F = 6.02$; df = 10, 120; $P < 0.01$). On empty shelves, the overall mean ± SEM percentage of parasitism, computed across all shelf levels, was 56.5 ± 3.0, 21.9 ± 3.0, and 23.1 ± 3.0% for *T. deion*, *T. ostriniae*, and *T. pretiosum*, respectively. The numerically highest percentage of parasitism occurred on shelf 4 for *T. deion* and shelf 3 for *T. ostriniae* and *T. pretiosum* (Fig. 2). On shelves with packages, the overall mean ± SEM percentage of parasitism was 60.1 ± 6, 16.6 ± 1.7, and 16.5 ± 3.1% for *T. deion*, *T. ostriniae*, and *T. pretiosum*, respectively, and the numerically highest percentage parasitism was observed on shelf 3 for all three species (Fig. 1). Percentage of parasitism for *T. deion* was similar on packaged and empty shelves, but it was significantly lower on packaged shelves for the other two species.

One-way ANOVAs for percentage of parasitism among shelf levels and within packaging treatment were not significant for *T. deion*, either on empty shelves or with packaging ($F = 2.37$; df = 5, 24; $P = 0.07$ and $F = 0.58$; df = 5, 24; $P = 0.72$, respectively), indicating an even distribution of parasitism among shelving levels (Table 1). However, significant differences were found for *T. ostriniae* on empty shelves; parasitism on shelf 3 was greater than shelves 1, 4, and 5, whereas parasitism on shelf 2 was greater than on shelf 5 ($F = 7.51$; df = 5, 24; $P < 0.01$) (Table 2). Percentage of parasitism also differed significantly among shelving levels for *T. ostriniae* on shelves with packaging. Parasitism was greater on the floor and shelf 3 than on shelves 1, 2, 4, or 5 ($F = 12.52$; df = 5, 24; $P < 0.01$) (Table 2). Similar trends in percentage of parasitism among shelves were
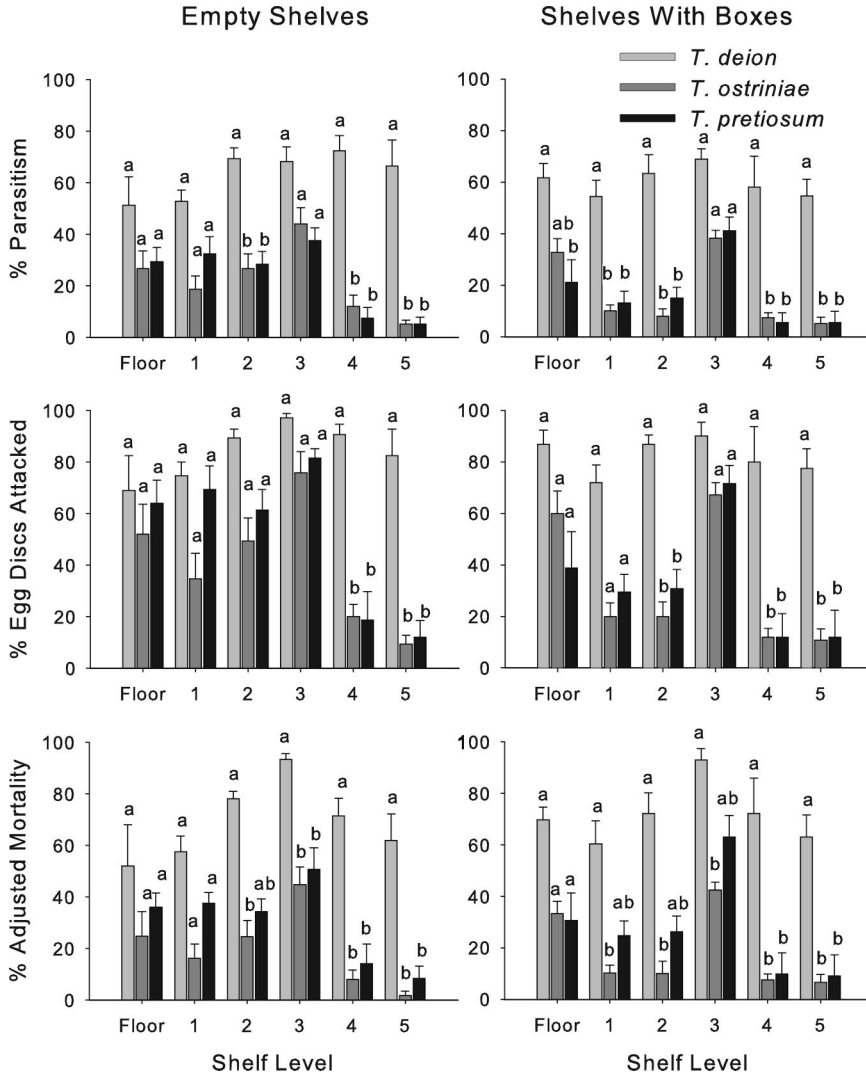
Fig. 2. Mean ± SEM percentages of egg parasitism, egg disks attacked, and corrected total mortality of *P. interpunctella* for three species of *Trichogramma* with empty shelves or shelves with packaging. Means within a shelf level with the same letter are not significantly different (Tukey–Kramer adjusted *t*-test of least-square means; *P* < 0.5).

observed for *T. pretiosum* on empty shelves, with the floor through shelf 3 significantly greater than shelves 4 and 5 ($F = 8.62$; df = 5, 24; $P < 0.01$) (Table 3). However, for *T. pretiosum* on shelves with packaging, the only significant differences observed were greater percentage of parasitism on shelf 3 compared with shelves 4 and 5 ($F = 5.92$; df = 5, 24; $P < 0.01$) (Table 3).

Mixed model *t*-tests by shelf and species for percentage of parasitism on empty shelves versus shelves with packaging were not significant at any shelf level for *T. deion*. In contrast, percentage of parasitism was significantly greater on empty shelves at shelf level 2 for *T. ostriniae* ($t = -2.98$, df = 8, $P = 0.02$) and at shelf level 1 for *T. pretiosum* ($t = -2.33$, df = 8, $P = 0.05$).

Percentage of Disks Attacked. Significant treatment effects and interactions for the percentage of disks

attacked (disks with at least one egg parasitized) included packaging ($F = 5.61$; df = 1, 24; $P = 0.03$), species ($F = 67.95$; df = 2, 120; $P < 0.01$), shelf level ($F = 27.12$; df = 5, 120; $P < 0.01$), and species × shelf ($F = 5.56$; df = 10, 120; $P < 0.01$). On empty shelves, the overall mean ± SEM percentage of egg disks parasitized across all shelf levels was 79.1 ± 2.9, 39.7 ± 4.7, and $50.8 \pm 2.4\%$ for *T. deion, T. ostriniae,* and *T. pretiosum,* respectively. The numerically highest percentage of egg disks parasitized was observed on shelf 3 for all three species (Fig. 2). On shelves with packages, the overall mean ± SEM percentage of egg disks parasitized across all shelf levels was 82 ± 5.4, 31.2 ± 2.2, and $31.9 \pm 5.3\%$ for *T. deion, T. ostriniae,* and *T. pretiosum,* respectively. Highest percentage of parasitism was observed on shelf 3 (parasitoid release point) for all three species (Fig. 2).

Table 1. Tukey's HSD multiple comparison for ONEWAY PROC GLM of percentage of parasitism, percentage of egg disks parasitized, and percentage of adjusted mortality for *T. deion* packaging treatments by shelf

	% parasitism	% egg disks parasitized	% corrected mortality
Empty shelves			
Floor	39.00 ± 10.08a	58.67 ± 13.47a	52.04 ± 16.00b
Shelf 1	50.67 ± 3.89a	73.33 ± 5.33a	57.46 ± 6.04b
Shelf 2	67.67 ± 3.10a	89.33 ± 3.40a	78.16 ± 2.70ab
Shelf 3	64.29 ± 5.84a	95.71 ± 1.75a	93.22 ± 2.43a
Shelf 4	64.00 ± 6.07a	82.67 ± 4.00a	71.45 ± 6.85ab
Shelf 5	54.00 ± 9.77a	76.00 ± 10.19a	61.83 ± 10.33ab
Shelves with packaging			
Floor	61.67 ± 5.55a	86.67 ± 5.58a	69.71 ± 4.96ab
Shelf 1	54.33 ± 6.25a	72.00 ± 6.80a	60.34 ± 8.85b
Shelf 2	63.33 ± 7.13a	86.67 ± 3.65a	72.13 ± 7.93ab
Shelf 3	68.93 ± 3.94a	90.00 ± 5.35a	92.81 ± 4.48a
Shelf 4	58.00 ± 12.07a	80.00 ± 13.50a	72.19 ± 13.67ab
Shelf 5	54.67 ± 6.46a	77.33 ± 7.77a	62.97 ± 8.62ab

Means ± SEM for each shelf level within a treatment with the same letter are not significantly different ($\alpha = 0.05$).

Table 2. Tukey's HSD multiple comparison for ONEWAY PROC GLM of percentage of parasitism, percentage of egg disks parasitized, and percentage of adjusted mortality for *T. ostriniae* packaging treatments by shelf

	% parasitism	% egg disks parasitized	% corrected mortality
Empty shelves			
Floor	26.67 ± 6.77ab	52.00 ± 11.62ab	24.64 ± 9.57abc
Shelf 1	18.67 ± 5.09bc	34.67 ± 9.75bc	16.17 ± 5.51abc
Shelf 2	26.67 ± 5.60ab	49.33 ± 8.84ab	24.48 ± 6.42ab
Shelf 3	43.93 ± 6.30a	75.71 ± 8.33a	44.75 ± 6.76a
Shelf 4	12.00 ± 4.33bc	20.00 ± 4.71bc	7.93 ± 3.70bc
Shelf 5	5.00 ± 1.58c	9.33 ± 3.40c	1.76 ± 1.50c
Shelves with packaging			
Floor	32.67 ± 5.36a	60.00 ± 8.69a	33.26 ± 4.81a
Shelf 1	10.00 ± 2.30b	20.00 ± 5.16b	10.27 ± 2.95b
Shelf 2	8.00 ± 2.76b	20.00 ± 5.58b	9.92 ± 4.88b
Shelf 3	38.21 ± 2.97a	67.14 ± 4.84a	42.43 ± 3.10a
Shelf 4	7.33 ± 1.94b	12.00 ± 3.27b	7.54 ± 2.29b
Shelf 5	5.00 ± 2.47b	10.67 ± 4.52b	6.57 ± 3.00b

Means ± SEM for each self level within a treatment with the same letter are not significantly different ($\alpha = 0.05$).

Table 3. Tukey's HSD multiple comparison for ONEWAY PROC GLM of percentage of parasitism, percentage of egg disks parasitized, and percentage of corrected mortality for *T. pretiosum* packaging treatments by shelf

	% eggs parasitized	% egg disks parasitized	% corrected mortality
Empty shelves			
Floor	29.33 ± 5.39a	64.00 ± 8.84a	35.94 ± 5.48ab
Shelf 1	32.33 ± 6.68a	69.33 ± 9.09a	37.41 ± 4.26ab
Shelf 2	28.33 ± 4.97a	61.33 ± 8.00a	34.29 ± 4.81ab
Shelf 3	37.50 ± 4.92a	81.43 ± 3.64a	50.56 ± 8.47a
Shelf 4	7.33 ± 4.30b	18.67 ± 11.04b	13.99 ± 7.74bc
Shelf 5	5.00 ± 2.74b	12.00 ± 6.46b	8.24 ± 4.80c
Shelves with packaging			
Floor	21.00 ± 8.77ab	38.67 ± 14.2ab	30.53 ± 10.68ab
Shelf 1	13.00 ± 4.58ab	29.33 ± 6.86ab	24.60 ± 5.71ab
Shelf 2	15.00 ± 4.15ab	30.67 ± 7.48ab	26.15 ± 6.17ab
Shelf 3	41.07 ± 5.33a	71.43 ± 7.14a	62.89 ± 8.51a
Shelf 4	5.33 ± 3.89b	12.00 ± 9.04b	9.84 ± 8.06b
Shelf 5	5.33 ± 4.55b	12.00 ± 10.41b	9.07 ± 8.14b

Means ± SEM for each self level within a treatment with the same letter are not significantly different ($\alpha = 0.05$).

One-way ANOVAs for percentage of disks attacked among shelf levels were not significant for *T. deion* either on empty shelves or shelves with packaging ($F = 2.37$; df = 5, 24; $P = 0.07$ and $F = 0.97$; df = 5, 24; $P = 0.72$, respectively) (Table 1). In contrast, significant differences were found for *T. ostriniae* on shelves without packages, with shelf 3 having a greater percentage of egg disks attacked than shelves 1, 4, or 5, and the floor and shelf 2 having a greater percentage of egg disks parasitized than shelf 5 ($F = 8.98$; df = 5, 24; $P < 0.01$) (Table 2). Similarly, on shelves with packages, *T. ostriniae* parasitized a greater percentage of egg disks on the floor and shelf 3 compared with shelves 1, 2, 4, or 5 ($F = 12.23$; df = 5, 24; $P < 0.01$) (Table 2). On shelves without packaging, *T. pretiosum* parasitized a greater percentage of egg disks on the floor and shelves 1, 2, and 3, compared with shelves 4 or 5 ($F = 11.32$; df = 5, 24; $P < 0.01$) (Table 3). However, the only significant differences for the percentage of egg disks parasitized by *T. pretiosum* among shelves with packaging were between shelf 3 and shelves 4 or 5 ($F = 5.7$; df = 5, 24; $P, < 0.01$) (Table 3).

Mixed model *t*-tests by shelf and species for percentage of disks parasitized on empty shelves versus shelves with packaging were not significantly different at any shelf level for *T. deion.* In contrast, percentage of egg disks parasitized was found to be significantly greater on empty shelves at shelf level two for *T. ostriniae* ($t = -2.55$, df = 8, $P = 0.03$) and at shelf levels 1 and 2 for *T. pretiosum* ($t = -3.53$, df = 8, $P < 0.01$ and $t = -2.78$, df = 8, $P = 0.02$, respectively).

Corrected total Percentage of Mortality. There were significant treatment effects and interactions for total corrected percentage of mortality with respect to shelf level ($F = 37.18$; df = 5, 120; $P < 0.01$), species ($F = 60.24$; df = 2, 24; $P < 0.01$), and species × shelf ($F = 4.13$; df = 10, 120; $P < 0.01$). On empty shelves, the overall mean ± SEM percentage of total mortality, computed across all shelf levels, was 68.7 ± 4.2, 19.7 ± 4.0, and 29.8 ± 2.7% for *T. deion*, *T. ostriniae*, and *T. pretiosum*, respectively, with the numerically highest percentage of total mortality occurring on shelf 3 for all three species (Fig. 2). On shelves with packages, the overall mean ± SEM corrected total percentage of mortality across all shelf levels was 71.5 ± 7.4, 18.1 ± 1.9, and 26.8 ± 5.7% for *T. deion*, *T. ostriniae*, and *T. pretiosum*, respectively, and the highest total mortality was observed on shelf 3 for all three species (Fig. 2).

One-way ANOVAs for corrected total percentage of mortality among shelf levels for *T. deion* were significantly different for both empty shelves as well as shelves with packaging. On empty shelves, egg mortality was greater on shelf 3 than on shelf 1 or the floor

($F = 3.72$; df $= 5, 24$; $P = 0.01$). On shelves with packaging, egg mortality was greater on shelf 3 than on shelf 1 ($F = 2.64$; df $= 5, 24$; $P = 0.05$) (Table 1). Significant differences also were found for *T. ostriniae* on empty shelves with egg mortality on shelf 3 greater than on shelves 4 or 5 ($F = 6.43$; df $= 5, 24$; $P < 0.01$) (Table 2). On shelves with packaging, *T. ostrinae* caused greater egg mortality on the floor and shelf 3 than on shelves 1, 2, 4, or 5 ($F = 9.95$; df $= 5, 24$; $P < 0.01$) (Table 2). Corrected total percentage of egg mortality was significantly different among empty shelves for *T. pretiosum*, with egg mortality on shelf 3 significantly greater than on shelves 4 or 5; whereas egg mortality on the floor and shelves 1 and 2 was significantly greater than on shelf 5 ($F = 7.66$; df $= 5$, 24; $P < 0.01$) (Table 3). On shelves with packaging, egg mortality due to *T. pretiosum* was found to be significantly greater on shelf 3 than shelves 4 or 5 ($F = 6.05$; df $= 5, 24$; $P < 0.01$) (Table 3).

Mixed model *t*-tests by shelf and species for corrected total percentage of egg mortality on empty shelves versus shelves with packaging were not significantly different at any shelf level for the three *Trichogramma* species.

Uniformity of Parasitism. On empty shelves, percentage of parasitism by *T. deion* was not uniformly distributed ($\chi^2 = 31.0$, df $= 5$, $P < 0.01$). However, the distribution of parasitism across shelves that had packages was uniform ($\chi^2 = 7.62$, df $= 5$, $P = 0.18$). For *T. ostriniae,* the distribution of percentage of parasitism across shelf levels was not uniform, either for empty ($\chi^2 = 186.2$, df $= 5$, $P < 0.01$) or shelves with packaging ($\chi^2 = 122.8$, df $= 5$, $P < 0.01$) shelving. Similarly, tests for *T. pretiosum* did not indicate a uniform distribution for the percentage of parasitism across shelves either for empty shelves ($\chi^2 = 153.9$, df $= 5$, $P < 0.01$) or shelves with packaging ($\chi^2 = 199.4$, df $= 5$, $P < 0.01$).

Discussion

Among the three species we compared, *T. deion* seems to have the best potential for use in augmentative release programs in retail stores and warehouses, based on dispersion of parasitism, parasitism rates, and parasitoid-induced host mortality. Percentage of parasitism of *P. interpunctella* eggs and total egg mortality caused by *T. deion* were ≈3 times greater than the other two species (Fig. 2). In addition, when *T. deion* was released at the vertical midpoint of the shelving (shelf 3), the pattern of parasitism was the most even, ranging from the floor level to the top shelf, suggesting that a central release point is likely to provide adequate vertical coverage. In contrast, parasitism by *T. ostriniae* and *T. pretiosum* was constrained to the release shelf or shelves below. In addition, parasitism by *T. ostriniae* and *T. pretiosum* rapidly diminished on shelves that were further below the release shelf. On shelves where all parasitoids found and used egg patches, the rate of egg disk use was similar among the three species. However, *T. deion* caused the highest overall mortality because it more completely attacked and parasitized eggs within a disk than *T. ostriniae* or *T. pretiosum* (Fig. 2).

Little information is available on the fate and the dispersal of egg parasitoids after release. In this study, *Trichogramma* spp. were most active around the release points. Presumably, the females are walking while foraging, resulting in a limited dispersal compared with flying parasitoids. This behavior is also advantageous in the stored-product context, because *Trichogramma* spp. are less likely to occur in areas of the stores where living insects are not tolerated. However, even different populations of one *Trichogramma* species may differ significantly in dispersal, as has been shown both in the laboratory and under semifield conditions for *T. deion* (Vereijssen et al., 1997, Silva, 1999) and for *Trichogramma cordubensis* Vargas & Cabello (Silva 1999). Vereijssen et al. (1997) observed that immediately after release, females of one strain of *T. deion* remained near the release site, whereas those of the other strain dispersed more rapidly.

It is interesting that on shelves without packaging, the percentage of parasitism by *T. deion* was highest above the release shelf, but overall mortality and patch use were highest on the release shelf. This may be caused by a density-dependent effect in which a greater ratio of parasitoids to eggs occurred, leading to greater egg patch discovery, accompanied by superparasitism, host killing, or both. Enhanced host mortality due to superparasitism or host killing would probably have minimal effects on successful Indianmeal moth control in retail stores and warehouses because future moth management will be dependent on regular augmentative releases rather than parasitoids cycling with the host.

Egg disks located on the floor were consistently parasitized at a rate similar to, or greater than, that for the first shelf among the three *Trichogramma* species. This was especially apparent for *T. ostriniae* and *T. pretiosum*, which foraged predominantly in the lower half of the shelving structure, and for which the floor was the second-most active level. Percentage of parasitism, disks used, and corrected total mortality on the floor were either similar to or significantly greater than the first or second shelves for all three species, and significantly greater than the fourth or fifth shelf for *T. ostriniae* and *T. pretiosum* (Fig. 2; Table 3). This suggests that individual *Trichogramma*, especially *T. ostriniae* and *T. pretiosum*, spent a disproportionate amount of time foraging on the floor, perhaps because individuals that leave the shelving unit are unlikely to return. It is thought that *Trichogramma* females find hosts within the host habitat by random search (Suverkropp 1997). Suverkropp (1997), studying dispersal of *Trichogramma brassicae* Bezdenko on maize, *Zea mays* L., found that flight was important in the movement from one plant to another, whereas walking was the most important method of movement within a plant.

Either negative photo- or geotaxis by *T. ostriniae* and *T. pretiosum* may explain the apparent absence of foraging on shelves above the release point. Other studies have documented patterns of vertical dispersal

in natural enemies, including *Trichogramma.* For example, *Trichogramma minutum* Riley foraging for the eggs of the spruce budworm, *Choristoneura fumiferana* (Clemens) (Lepidoptera: Tortricidae), was found to preferentially attack greater numbers of egg clusters when released at 0.25 m, but lower number of clusters when released at ground level (Smith 1988). Comparing the vertical dispersal of *T. deion* and *T. cordubensis* on tomato (*Lycopersicum* spp.) plants, Silva (1999) found one population of *T. cordubensis* that parasitized significantly more egg masses higher on the plant.

Packaging did not have a significant effect on the dispersion of parasitism by *T. deion*, but it did affect both the percentage of eggs parasitized and the percentage of egg disks attacked by *T. ostriniae* and *T. pretiosum.* Differences between empty shelves and those with packaging were restricted to levels below the release point. One reason why packaging did not significantly affect parasitism is that the spatial scale at which *Trichogramma* interact with their environment is much finer than the scale at which packaging adds complexity to it. Experiments designed to explore the impact of finer scale habitat complexity, including loose flour and millet, on the three *Trichogramma* species, showed that these finer scale features did significantly affect host-foraging success (Grieshop 2005). In addition, a study by Schöller et al. (1996) showed that *Trichogramma* foraging in bulk grain were unable to locate sentinel egg patches buried more than a few centimeters. In a study comparing the response of parasitoids of varying body sizes responding to habitat patches of different grain and extent, Roland and Taylor (1997) demonstrated the importance of the spatial scale at which parasitoids interact with their habitat. In their study, the smallest parasitoids of the forest tent caterpillar were affected only by fine-grain habitat fragmentation and not by fragmentation taking place at broader spatial scales.

In conclusion, among the three species tested, *T. deion* seems to be the best-suited species for inundative releases in retail store and warehouse environments. *T. deion* parasitized a greater percentage of eggs on almost all shelving levels and had the most even vertical pattern of parasitism. For augmentative biological control programs in retail warehouse settings, *T. deion* would require fewer releases than the other two species tested because of its ability to disperse and find hosts. This would result in a cost savings for the manager because fewer *Trichogramma* would need to be purchased, and fewer release points would result in reduced labor costs.

Acknowledgments

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